Available online at www.sciencedirect.com
ScienceDirect
Colloids and Surfaces A: Physicochem. Eng. Aspects 299 (2007) 133–145
COLLOIDS AND SURFACES A
www.elsevier.com/locate/colsurfa

Liquid micellar discontinuous cubic mesophase from ternary monoolein/ethanol/water mixtures

Rivka Efrat [a,1], Abraham Aserin [a], Ellina Kesselman [b], Dganit Danino [b], Ellen J. Wachtel [c], Nissim Garti [a,*]

[a] *Casali Institute of Applied Chemistry, The Institute of Chemistry, The Hebrew University of Jerusalem, Jerusalem 91904, Israel*
[b] *Department of Biotechnology and Food Engineering, Technion-Israel Institute of Technology, Haifa 32000, Israel*
[c] *Faculty of Chemistry, The Weizmann Institute of Science, Rehovot 76100, Israel*

Received 27 September 2006; received in revised form 12 November 2006; accepted 14 November 2006
Available online 19 November 2006

Abstract

In constructing the phase diagram of glycerol monooleate (GMO)/ethanol/water mixtures we detected, in addition to the well-characterized regions of lamellar and cubic bicontinuous liquid crystals, three additional isotropic transparent regions that were not previously well-defined: a large isotropic region denoted in early studies as the L_1-phase, an isotropic region with characteristics similar to the sponge L_3-phase, and a third isotropic region which had not been characterized previously. The present study deals with efforts to characterize this unique isotropic region. The isotropic region is an "island" located within a two phase region connecting the three isotropic regions of the cubic, lamellar, and micellar phases. We termed this phase I_2 (Q_L) phase or the Q_L phase, which stands for cubic liquid mesophase. This phase is a totally transparent and non-birefringent fluid (completely pourable). It is stable even after over 9 months of storage at room temperature. The fluid is of low viscosity and almost Newtonian. The new phase shows many characteristic features of an ordered phase. Small-angle X-ray scattering measurements (SAXS), SAXS-synchrotron temperature-dependent measurements, and cryogenic-transmission electron microscopy (cryo-TEM) observation reveal that the phase has a cubic symmetry. The SAXS diffractions indicate formation of a somewhat less ordered discontinuous cubic micellar mesophase. The cryo-TEM images provide strong evidence for ordered domains with cubic symmetry. Complementary measurements confirm that the phase is a unique case of a micellar cubic phase. We expect that the fluidic ordered mesostructure will have future applications as a liquid vehicle for the solubilization of nutraceuticals, cosmetoceuticals, and pharmaceuticals.

Keywords: Phase diagrams; Cubic mesophases; Cryo-TEM; SAXS; Electrical conductivity; Phase transitions; Ethanol effect; Glycerol monooleate

1. Introduction

A large variety of amphiphilic molecules in polar solvents (usually water) create mesophases. The most common mesophases are: lamellar (L_α,), which consist of parallel stacks of surfactant bilayers, hexagonal (H_1 and H_2—normal and reverse hexagonal symmetries), and cubic phases (Q phases) which display three-dimensional long-range order. Cubic phases can consist of bicontinuous interpenetrating networks of solvent and amphiphile (V-phases) and, in some particular cases, consist of discrete micellar aggregates (I-phases). In addition, the cubic phases can be normal (I_1 and V_1) or inverted (I_2 and V_2) [1–3]. The bicontinuous cubic phases have been extensively studied while the discontinuous phases are only rarely detected and almost unstudied.

Cubic bicontinuous mesophases exhibit the most complex spatial organization of all known lyotropic liquid crystals (LLC). They are very viscous, nearly solid in some cases (ca. 10^6 Pa), and are optically isotropic. The cubic phases consist of two separate, continuous but non-interacting hydrophilic regions divided by a surfactant bilayer that is contorted into an infinite periodic minimal surface (IPMS). IPMS have zero mean curvature, and when the Gaussian curvature (K) is negative the surface is hyperbolic [4–6]. The three common IPMS surfaces relevant to lipid–water systems are the P-primitive surface (Schwartz primitive, Q^{229} surface or $Im3m$ space group), the D-diamond surface

* Corresponding author.
E-mail address: garti@vms.huji.ac.il (N. Garti).
[1] Part of the results presented in this paper has been included in the dissertation of R.E. for the degree of Doctor in Applied Chemistry at The Hebrew University of Jerusalem, Israel.

0927-7757/$ – see front matter © 2006 Elsevier B.V. All rights reserved.
doi:10.1016/j.colsurfa.2006.11.029

(Schwartz diamond, Q^{224} F-surface or *Pn*3*m* space group), and the G-gyroid surface (Shoen's gyroid, Q^{230}, or *Ia*3*d* space group) [5,7].

It has been suggested that cubic micellar mesophases (I_1-phase) exist in three different packing forms: simple cubic, body centered cubic (bcc), and face centered cubic (fcc) [8,9]. Luzzati et al. and Burducea have suggested that the most common form is closed-packed in bcc mode and consists of identical quasi-spherical micelles (symmetry *m*3*m*) with an *Im*3*m* (Q^{229}) space group. The two other cubic phases are apparently composed of two types of micelles with different symmetry, the *Fd*3*m* and *Pm*3*n* space groups. The *Fd*3*m* (Q^{227}) space group is closed-packed in fcc symmetry and consists of two types of quasi-spherical micelles, one with $\bar{3}m$ symmetry, and the second with $\bar{4}3m$. The *Pm*3*n* (Q^{223}) space group contains quasi-spherical micelles with 3 m symmetry, and slightly flattened micelles with $\bar{4}2m$ symmetry [10]. Hyde [11] has suggested that the discrete micellar mesophases of type 2 systems can form *Fm*3*m* or $P6_3/mmc$, *Fd*3*m*, *Im*3*m*, and *Pm*3*n* symmetries.

It has been suggested that additional exotic lyotropic mesophases, such as polycontinuous sponges, meshes (both smectic and 3D crystals), and a novel 3D columnar packing, are spontaneously formed [12,13]. Recent developments in theoretical and experimental studies of possible intermediate mesophases revealed formation of phases with novel geometries and topologies, such as a cubic array with infinite branched bilayer topologies, and an Archimedean screw with minimal tricontinuous surface (with symmetry $I4_132$) [12].

The transitions between liquid crystalline phases include topological rearrangements of the bilayer, and extensive creation or rupture of connections between surfactant/water interfaces. It has frequently been suggested that transitions to non-lamellar phases occur via the formation of intermediate structures. Siegel [14] has developed a model for such transitions occurring in several stages. The model includes fusion of the membrane, known as a stalk, and radial expansion into smaller structures known as hemifusion intermediates or transmonolayer contacts (TMCs). TMCs play an important role in the lamellar (L_α)/inverted hexagonal (H_{II}) phase transition, or, alternatively, they can rupture to form fusion pores called interlamellar attachments (ILAs). ILAs are structural elements that are precursors of Q_{II} phases [14].

It should be stressed that different phases form as a result of the interplay, or competition, between local interactions and global constraints or geometrical (topological) rules imposed by space filling requirements [5,15]. Two major approaches provide useful insight into the self-assembly processes in amphiphilic systems: the steric packing model, and the surface-bending model [2,12,16–18].

Addition of a third component to binary systems of lyotropic liquid crystals can alter the factors responsible for phase stability and can cause phase transitions. Transverse interactions such as hydration between bilayers can be disproportionately modified by forming lipid mixtures. Lateral interactions could also be modified in a non-additive way, with a strong effect on phase stability [5,19,20]. Phases of non-uniform interfacial curvature can become favored by partial lateral segregation of different lipid species into regions of different curvature. Regions of two phase coexistence might become more extensive, and three-phase coexistence may become possible. Finally, new phases which do not appear in purely binary lipid–water systems could form.

Several phase diagrams of three component mixtures of glycerol monooleate (GMO) with various polyols and hydrotropes were reported [21–23]. Engström et al. added polar solvents such as DMSO, propylene glycol, PEG 400, and ethanol to GMO/water mixtures, and constructed ternary phase diagrams [21]. In these systems an L_3 (sponge) phase appeared, while it is absent in the binary GMO/water system. These investigations focused mainly on the formation of major mesophases, such as cubic, lamellar, and hexagonal, and regions outside the borders (including the L_3-phase) of these regions were less studied.

In our studies we are interested in identifying intermediate mesophases with specific physical properties and characteristics. This report will deal with the effect of added ethanol on the reorganization and transformations occurring in bicontinuous cubic phase and micellar phase in the presence of increasing amounts of water and ethanol.

In the course of reconstructing the phase diagram of GMO/ethanol/water, we located a small island of an isotropic phase (within the two phase region) in the vicinity of the cubic phase and not far from the border line of the micellar structures that display interesting and unique structural and physical characteristics. The focus of this paper is the structural characterization of this unique mesophase. We termed this phase a Q_L cubic liquid. The characterization of this small one phase region is interesting from both the scientific and application point of view since it is a non-viscous, transparent fluid (and not a gel) with long-range order. This phase is ideal for the formation of dispersed particles known as cubosomes. The phase has also unique solubilization capacities of bioactive molecules.

2. Experimental

2.1. Materials

Monoolein (distilled glycerol monooleate, GMO) that consists of 97.1 wt% monoglyceride and 2.9 wt% diglyceride (acid value 1.2, iodine value 68.0, melting point 37.5 °C, and free glycerol 0.4%) was obtained from Riken Vitamin Co. Ltd., Japan. Ethanol was analytical reagent (>99%) and purchased from Frutarom Ltd. (Israel). Water was double distilled.

2.2. Phase diagram

The ternary phase diagram was constructed from GMO/ethanol/water mixtures at 25 ± 0.5 °C. Mixtures of GMO/water were prepared in culture tubes sealed with Viton-lined screw caps in predetermined weight ratios. The phase diagrams were constructed by gradual titration of the GMO/water mixtures with ethanol. The samples were allowed to equilibrate for at least 24 h and an extra 1 wt% of ethanol was added to each sample to guarantee they would reach phase equilibrium. The identification and characterization of the different phases in the prepared samples, along the dilution lines, was done using